NOTES ON W. F. K. STOCK'S NITROGEN PROCESS.

By W. Pearson Skertchly.

In The Analyst, vol. xvii., No. 194, appeared a paper by Mr. W. F. K. Stock on a "New and Rapid Method for the Determination of Nitrogen in Organic Bodies." The process

is based upon the oxidation of the substance by a mixture of strong sulphuric acid and manganese dioxide. This paper was read before a meeting of the members of the Society in May last, and, from the discussion which followed the reading of the paper, it appears that several members cast considerable doubt on the accuracy of the method, fearing that the use of manganese dioxide would cause considerable error in the estimation of the nitrogen.

Mr. Stock wrote a supplementary paper (THE ANALYST, xvii., 196), to prove that there was not the slightest danger of losing any nitrogen if the process was properly carried out.

I have also made a series of experiments to test the efficacy of the method, both with pure ammonia salts and with various nitrogenous organic substances. The figures that I obtained in my analyses I now wish to bring before you.

A solution of crystallized ammonium sulphate was made up, and two separate distillations gave 0·15895 grm. and 0·15810 grm. as the ammonia (NH_3) contents of 20 c.c. of this solution.

20 c.c. of the solution were now evaporated to dryness, with the addition of a few drops of dilute sulphuric acid, and treated with 10 c.c. of strong sulphuric acid and 5 grms. of manganese dioxide, in the way described by Mr. Stock, heating the mixture for ten minutes on a sand-bath. On distillation I obtained 0·15970 grm. NH_3 or 100·7 per cent. of the quantity taken.

Another 20 c.c. of the solution were treated exactly the same as in the above experiment, except that the heating on the sand-bath was continued for three-quarters of an hour. I recovered 0·15844 grm. NH_3 or 99·95 per cent.

The same quantity of the solution was then evaporated, and to it was added 1 grm. of pure cane sugar. It was now heated with 15 c.c. of strong sulphuric acid and 7 grms. of manganese dioxide. A dark green colour appeared after heating for fifteen minutes. The ammonia was distilled off, and gave 0·15923 grm. NH_3, or 100·4 per cent.

As the amount of ammonia taken was so small, I think that these results are quite passable.

A sample of Dog-biscuit was analysed by this method, and gave the following results:—

Kjeldahl.	*New Method.*
3·43 per cent. Nitrogen.	3·51 per cent. Nitrogen.
4·16 ,, Ammonia.	4·26 ,, Ammonia.

A sample of Cotton-meal gave good results:—

Kjeldahl.	*New Method.*
4·01 per cent. Nitrogen.	3·94 per cent. Nitrogen.
4·87 ,, Ammonia.	4·78 ,, Ammonia.

Wheat-flour is very easily analysed by this method, as it is in such a fine state of division. The results obtained were :—

Kjeldahl.	*New Method.*
1·73 per cent. Nitrogen.	1·72 per cent. Nitrogen.
2·10 „ Ammonia.	2·09 „ Ammonia.

Coffee is rather difficult to analyse, owing to an excessive amount of frothing when heated even gently with sulphuric acid and manganese dioxide, and also because some of the pieces of coffee are carried up the sides of the beaker by the froth, and remain unacted upon. This latter difficulty, of course, may be got over by reducing the coffee to a very fine powder. I find that the frothing is to a great extent avoided by heating the 2 grm. of the sample with 20 c.c. of concentrated sulphuric acid alone, and then, when the coffee is completely dissolved, cautiously adding the manganese dioxide. The mixture now simply requires to be left on the sand-bath until a green colour distinctly appears.

A sample of Coffee gave the following results :—

Kjeldahl.	*New Method.*
2·30 per cent. Nitrogen.	2·23 per cent. Nitrogen.
2·79 „ Ammonia.	2·70 „ Ammonia.

A sample of commercially pure Gelatin gave satisfactory results :—

Kjeldahl.	*New Method.*
15·68 per cent. Nitrogen.	15·57 per cent. Nitrogen.
19·04 „ Ammonia.	18·90 „ Ammonia.

I now proceeded to analyse six samples, rich in nitrogen, kindly given to me by Dr. Bernard Dyer, and in which he had estimated the nitrogen by the Kjeldahl method.

The following is a list of the substances examined, and the percentages of nitrogen (and ammonia) found in each, by Dr. Dyer and myself :—

	New Method.		*Kjeldahl.*		*Dr. Dyer's figures (Kjeldahl.)*	
	Nitrogen.	Ammonia.	Nitrogen.	Ammonia.	Nitrogen.	Ammonia.
Linseed Cake—	3·49 p.c.	4·25 p.c.	3·87 p.c.	4·70 p.c.	3·91 p.c.	4·74 p.c.
Crushed Hoofs and Horns—	13·37 p.c.	16·23 p.c.	15·34 p.c.	18·62 p.c.	15·38 p.c.	18·68 p.c.
	13·30 p.c.	16·15 p.c.				
Fish Guano (1)—	7·80 p.c.	9·47 p.c.	9·27 p.c.	11·25 p.c.	9·24 p.c.	11·22 p.c.
	7·72 p.c.	9·37 p.c.				

Fish Guano (2)—					
4·94 p.c.	6·00 p.c.	5·50 p.c.	6·68 p.c.	5·60 p.c.	6·80 p.c.
4·97 p.c.	6·03 p.c.				
Dried Blood—					
10·81 p.c.	13·12 p.c.	12·93 p.c.	15·65 p.c.	13·08 p.c.	15·88 p.c.
10·82 p.c.	13·14 p.c.				
Manure—					
5·56 p.c.	6·75 p.c.	6·16 p.c.	7·49 p.c.	6·09 p.c.	7·40 p.c.

Finally, a sample of pure ammonium sulphate was taken, which should contain, theoretically, 21·21 per cent. nitrogen, and on distilling the ammonia out of a known quantity I obtained 21·25 per cent. nitrogen. On treating another portion by the "Stock Method" I found 21·05 per cent. nitrogen. This shows that my standard solutions used in the analyses were practically correct.

On looking over the last six substances given above, one will observe that the higher the percentage of nitrogen the greater is the difference between the "Stock" figure and the Kjeldahl figure. However, the loss is not proportionate to the amount of nitrogen present, and, as will be seen from the following table, the percentage loss of nitrogen is by no means the same. Thus, in

The Linseed Cake	the loss is	10·28	per cent	of the total	nitrogen present.
Hoofs and Horns	,,	13·15	,,	,,	,,
Fish Guano (1)	,,	17·28	,,	,,	,,
,, (2)	,,	10·63	,,	,,	,,
Dried Blood	,,	16·84	,,	,,	,,
Manure	,,	9·30	,,	,,	,,

The percentage of nitrogen found by the Kjeldahl process is taken as being the true percentage of nitrogen in the substances examined. There are several possible explanations of the deficiency, and I will enumerate a few of these.

(i.) The nitrogen in the presence of so much oxidizing agent may be oxidized to nitric acid straight away, and so be driven off.

(ii.) The ammonium sulphate formed may be further oxidized into nitrogen itself, or into some volatile compound of nitrogen.

(iii.) There may exist two different nitrogenous components in the above substances, one of which yields up its nitrogen, forming ammonium sulphate, while the other nitrogen is driven off by the strongly oxidizing mixture.

(iv.) The loss may result from some defect in the manipulation of the analyses, and this is, to some extent, borne out by the analyses I have given above. In the first experiments made it will be seen that the figures obtained

by both methods agreed fairly well with each other. It so happens that in these early analyses the manganese dioxide was added slowly, a little at a time, until I found out how much was required to quickly complete the oxidation and produce the green colour, which Mr. Stock says is an indication that the reaction is complete.

As soon as the requisite quantity was ascertained, it was added bodily to the substances, of which I afterwards give the figures. It is conceivable that in this case the large amount of oxygen present at one time was too much for the nitrogen, and that it is driven off, while in my former experiments the gradual addition of the manganese dioxide may only have produced sufficient oxygen to oxidize the sulphurous acid formed as the heating went on. When the substance was decomposed the green colour appeared, and no more dioxide was added. This part of the process requires further investigation, and I intend to do some more work on it.

I presume that a mixture of sulphuric acid and manganese dioxide acts on nitrogenous organic substances in much the same way as a mixture of sulphuric acid and potassium permanganate. It is known that the addition of potassium permanganate to a "Kjeldahl," even when all the organic matter is decomposed and the solution is colourless, may cause a considerable loss of ammonia.

In my experiments it always takes about ten minutes to a quarter of an hour to oxidize the substance. Mr. Stock says that he can carry out the operation in three to five minutes. Probably the difference in time required is due to the purity or impurity of the manganese dioxide used. Mine contains 40·86 per cent. of the dioxide. It is ordinary commercial powdered dioxide.

From the above experiments it is evident that the method requires a great deal more examination, and the cause of the loss of nitrogen discovering, together with, if possible, a preventative against the said loss, before the process can be used by analysts as a reliable one for the estimation of nitrogen in common organic substances.

Discussion.

Mr. F. H. Perry Coste remarked, with regard to the results obtained by Mr. Skertchly, that in view of the very good correspondence between the figures in several instances, and also of the results on pure salts recently communicated by Mr. Stock himself, he did not attach much importance to the low nitrogen result with the ammonium sulphate; he had very little doubt that that was due to some oversight in working, and that Mr. Skertchly on repeating the experiments would obtain the full nitrogen.

The differences in the case of such samples as the *Hoofs and Horns* were, however, very important, for these might be taken as criteria; and any method that would yield the full nitrogen with such materials as these might safely be accepted as reliable for all ordinary work. Of course the differences were so great as to make it clear that the

Stock method was useless as a general analytical process at present; but no one who had had personal experience in working out the Kjeldahl process would be hasty in finally condemning this new process on such grounds. It required long experimenting to bring the Kjeldahl process into thoroughly reliable working order, and he well remembered that for many weeks results were obtained regularly too low, very much as with the Stock method in the present case. As to the cause of the error here, having regard to the concordance between Mr. Skertchly's duplicates, he did not think it likely that the low nitrogen was due to any error in working; the results seemed to him too constant for that. Two sources of error were possible. The whole of the nitrogen might be converted into ammonia, and then some of it oxidised into a nitrogen oxide by the continued action of the manganese oxide, just as he had found that a heavy loss of nitrogen might occur in the Kjeldahl process if, at the end of the digestion, permanganate were added and the digestion renewed. This was a *time reaction*, for instance, he (Mr. Coste) had proved in Dr. Dyer's laboratory several years ago that if pure ammonium sulphate sulphuric acid and mercury were boiled with potassium permanganate for increasing periods of time, increased losses of nitrogen took place. Now, if Mr. Skertchly would treat these samples by the Stock method for various times (say 5 to 10 or 15 minutes) then, if he found increasing losses of nitrogen, that would point to a similar danger. If, however, in such experiments the nitrogen remained constant though still too low, that would strongly point to the second possible cause of error being present, viz., that the nitrogen in such samples exists in at least two different forms, and that the Stock process fails to convert the whole. Considering the nature of these samples, that explanation had frequently suggested itself to Dr. Dyer and himself; and this would account both for the agreement between Mr. Skertchly's duplicate "Stocks," though both so low, and also for the correct results obtained from other samples of a similar character. He hoped that Mr. Skertchly would continue these experiments, which were of great interest and value.

Dr. Bernard Dyer said that he himself had not yet found time to make any experiments with Mr. Stock's process. He was very anxious to get some personal experience of the process, and had been very pleased to give Mr. Skertchly those samples, so that by comparing results, he was enabled to get some indirect personal experience of what results the Stock process gave on materials of which he knew the composition.

The President wished to ask Mr. Skertchly whether he followed exactly the plan proposed by Mr. Stock, or in what way he altered it? Did he use the same quantities which Mr. Stock proposed, and did Mr. Stock lay down any rules as to the adding of manganese dioxide?

Mr. Coste assumed that in all these cases the distillations after the digestion in the Stock and Kjeldahl processes were carried out under equal conditions as to time and everything else.

Dr. Dyer, in reply to the question of a member, said that he had found no difficulty in obtaining acid which yielded practically no nitrogen. When a new bottle of acid was opened or a stock of caustic soda was made up, a complete blank experiment was made, going through the whole process, except that no nitrogenous substance was used, and the total allowance indicated for error and impurities from all sources came well within half a milligramme of nitrogen. There was no difficulty in getting sulphuric acid as pure as this.

Mr. Skertchly, in reply, said that as Mr. Stock had only used such small quantities, namely 0·5 to 1·0 gramme of the substances for analysis, he had thought it better to take somewhat larger amounts, up to 2 grammes. He first tried the process using the smaller quantity and the results thus obtained did not agree so well between themselves as when he used 2 grammes of the substance. Two grammes would give a better average sample than 0·5 gramme. With the larger amounts double the quantities of sulphuric acid and manganese dioxide were used. As to Mr. Coste's question, he would like to say that he gave one experiment with ammonium sulphate 10 minutes, and another three-quarters of an hour heating on the sand bath, and obtained the same results in both cases. With regard to the distillations, they were carried out under practically the same conditions.

He wished to thank Mr. Hehner for his kind assistance during the investigation.

The President said he was very glad to see that the Society had in Mr. Skertchly a new acquisition to those who read papers. It was highly desirable that young chemists, who became members, should attack matters requiring investigation, and give the Society the benefit of their experience.

Mr. Richmond read papers "On the action of some Enzymes on Milk-Sugar," and "The Estimation of Total Solids in Milk." With this the proceedings of the Society terminated.